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Lyophyllum rhombisporum sp. nov. from China

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Abstract — A new species, collected from Yunnan Province, China, is here described as *Lyophyllum rhombisporum* (*Lyophyllaceae, Agaricales*). The new species is macroscopically and microscopically very similar to *L. sykosporum* and *L. transforme*, but its spore shape is quite different. Molecular analysis also supports *L. rhombisporum* as an independent species.

Key words — *Agaricomycetes*, mushroom, taxonomy, ITS, phylogeny

Introduction

Lyophyllum is a genus in the *Lyophyllaceae* (*Agaricales*) with about 40 species, which are widespread in north temperate regions (Kirk et al. 2008). A few *Lyophyllum* species are well-known edible mushrooms, such as *L. shimeji* (Kawam.) Hongo, *L. decastes* (Fr.) Singer, and *L. fumosum* (Pers.) P.D. Orton. Research in China has reported 19 *Lyophyllum* species (Li 2002; Li et al. 2005; Dai & Tolgor 2007; Li & Li 2009), but more research is still needed on the taxonomy, molecular phylogeny, and cultivation of this genus. Here we describe a new species, *L. rhombisporum* from Yiliang County, Yunnan Province, in southwestern China.

Materials & methods

Microscopic and macroscopic characters were described based on the specimen material (L1736) following the methods of Yang & Zhang (2003). Sections were made by hand with a razor blade, then mounted in 5% KOH solution and lactophenol cotton blue respectively under an Olympus BH-2. The specimens were deposited in the Herbarium of Yunnan Academy of Agricultural Sciences, Kunming, China (YAAS).

Total DNA was extracted using the modified CTAB methodology (Hofstetter et al. 2002; Li et al. 2011). The internal transcribed spacer region (ITS) was amplified using

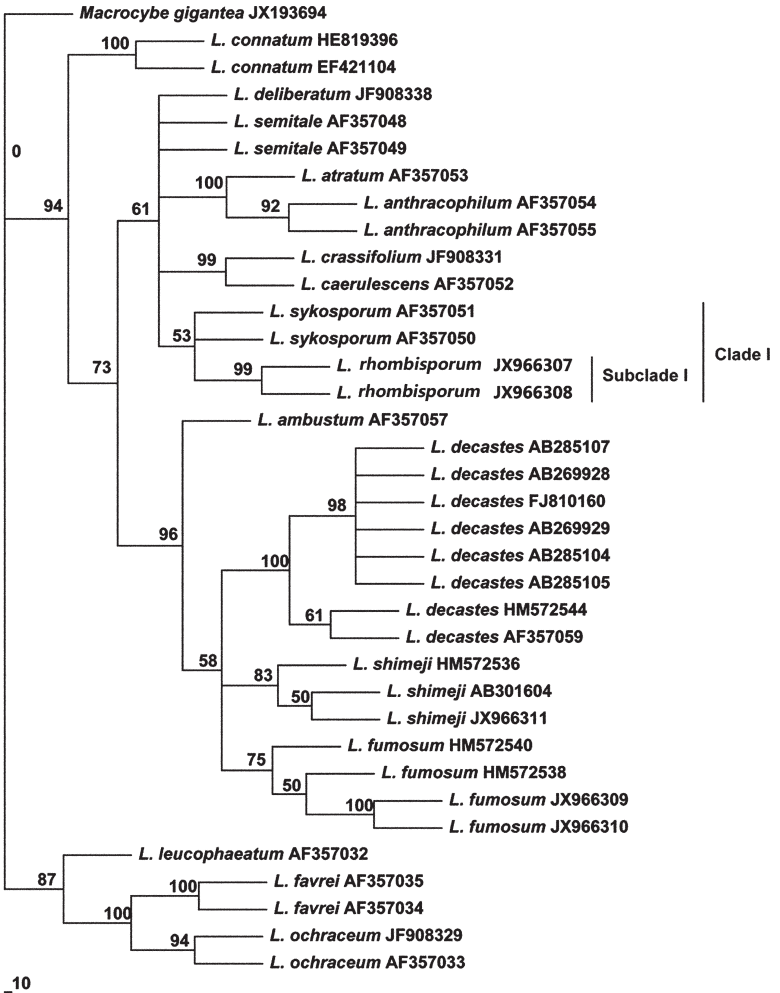

Plate 1. Equally weighted parsimony tree of the genus *Lyophyllum* based on ITS sequences from 36 ITS sequences. Numbers above branches indicate bootstrap support.

primers ITS4 and ITS5 (Hofstetter et al. 2002; Larsson & Sundberg 2011), following the modified Li's procedure (Li et al. 2011). Purifying and sequencing of PCR products were conducted by Beijing Genomics Institute.

Thirty-six sequences were included in molecular analysis: six generated from this research and another 30 retrieved from GenBank. *Macrocybe gigantea* (Massee) Pegler & Lodge was selected as the outgroup. Sequences were edited and assembled using SeqMan II (Larsson & Sundberg 2011), aligned by Muscle 3.7, and adjusted manually using BioEdit 7.0.1. Phylogenetic analyses were carried out via PAUP* 4.0 BEAT. Equally

weighted parsimony (MP) was used to search for optimal trees according to Hofstetter et al. (2002) and Larsson & Sundberg (2011).

Results & discussion

Phylogenetic analysis

Six strict consensus trees were generated based on the MP analyses of 36 ITS sequences (L = 479, CI = 0.714, RI = 0.866, RC = 0.618) (Plate 1). The phylogeny supports *L. rhombisporum* as distinct from the sampled *Lyophyllum* species, although it is closely related to *L. sykosporum*. In the strict consensus tree, *L. rhombisporum* and *L. sykosporum* form Clade I with 53% bootstrap support. This subdivides into two subclades, with Subclade I comprising *L. rhombisporum* with a strong bootstrap support (bs = 99%) and separated from Subclade II comprising *L. sykosporum*.

Taxonomy

Plate 2. Fruit-body of *Lyophyllum rhombisporum*.

Lyophyllum rhombisporum Shu H. Li & Y.C. Zhao **sp. nov.** Plates 2, 3

MycoBank MB 801752

Differs from *Lyophyllum sykosporum* and *L. transforme* by its rhombic basidiospores.

Type: China: Yunnan, Yiliang County, 24°42′N 103°09′E, alt. 1922 m, in forest of *Pinus yunnanensis* Franch, 23.6.2011, Shuhong Li, (**Holotype**, YAAS L1763; GenBank JX966307).

Etymology: Latin *rhombisporum*, referring to the rhombic or subrhombic shape of the spores.

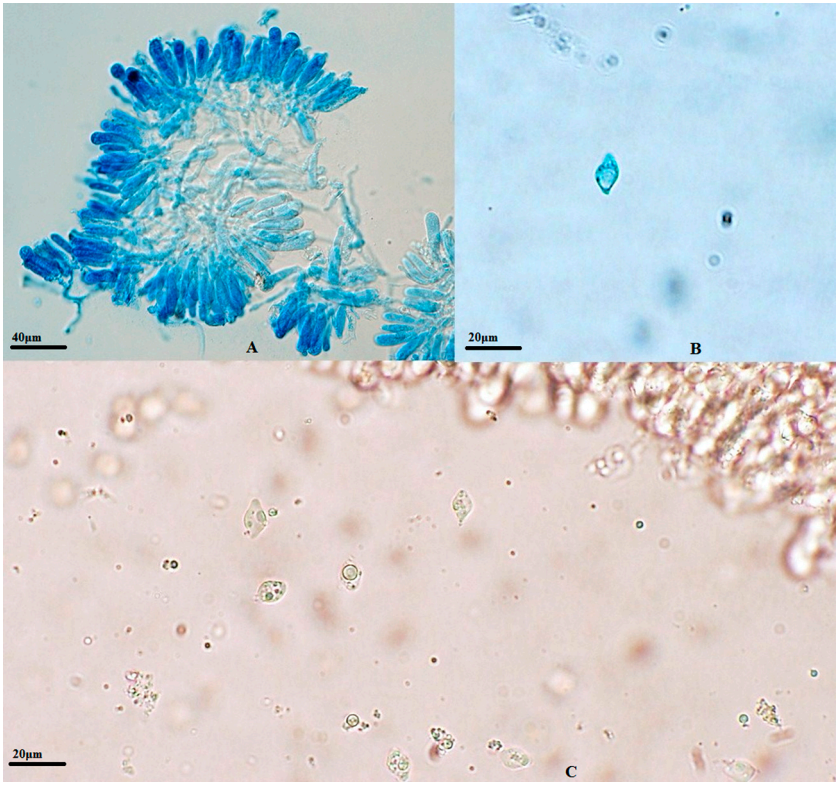

Plate 3. *Lyophyllum rhombisporum* (Holotype L1763). Fig. A. Basidia in lactophenol cotton blue; Fig. B. Spores in 5% KOH; Fig. C. Spore in lactophenol cotton blue.

Pileus 2.0–7.5 cm, convex, applanate at maturity, smooth, dry, not striate, greyish to pale taupe. Context thin, white to whitish grey. Odor and taste not distinctive. Stipe 3.0–8.0 long, 0.6–1.5 cm thick, cylindrical, greyish or greyish brown, slightly enlarged downwards, appressed fibrillose, pale brown at base. Lamellae moderately close together, adnate or subdecurrent, broad, whitish to greyish, blackening when bruised, edge blunt). Hymenophoral trama regular, consisting of hyaline hyphae, clamp-connections rare. Spores rhombic or subrhombic, hyaline, cyanophilous, with a single large oil-droplet or a few small oil-droplets, smooth, 14.5–17 × (7.5–)10–11.5 μm, Qm = 1.3. Basidia 36.5–46 × 10–11.3 μm, 4-spored, clavate, with siderophilous granulation well visible in lactophenol cotton blue or cotton blue, clamp connections absent. Pleuro- and cheilocystidia absent.

Ecology & Distribution: Scattered or gregarious in forests of *Pinus yunnanensis*. Summer to autumn. Known only from Yunnan Province, China.

Additional specimen examined: **CHINA**: **Yunnan**, Yiliang County, 24°42′N 103°09′E, alt. 1952 m, in *Pinus yunnanensis* forest, 21.9.2011, Shuhong Li, (YAAS L2082; GenBank JX966308).

Comments — This species is characterized by large sub-rhombic or rhombic basidiospores, and white or greyish gills that blacken when bruised, a feature of section *Lyophyllum* (Breitenbach & Kränzlin 1991; Hofstetter et al. 2002; Larsson & Sundberg 2011). Morphologically *Lyophyllum rhombisporum* is morphologically similar to *L. sykosporum* Hongo & Clémençon and *L. transforme* (Sacc.) Singer (Breitenbach & Kränzlin 1991; Hofstetter et al. 2002), which are distinguished by their triangular basidiospores. The phylogenetic analysis supports *L. rhombisporum* as a new species.

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